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None
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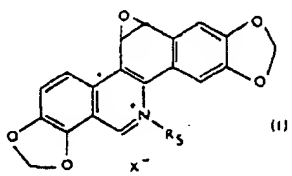
C2C
(71) Applicant
Quatrum-Empresa Nacional De Quimica Organica SARL. (Portugal),
6th Floor (right),
10 Avenida Joao XXI,
1000 Lisboa,
Portugal
(72) Inventors
Jose Manual Gasper Pereira,
Joaquim da Rocha Madureira
(74) Agent and/or address for service
Frank B. Dehn and Co.,
Imperial House,
15—19 Kingsway,
London,
WC2B 6UZ

(54) **Chemical compounds**

(57) Compounds of formulae I—VI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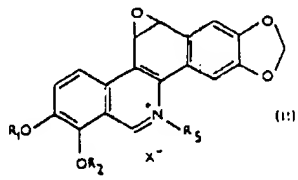
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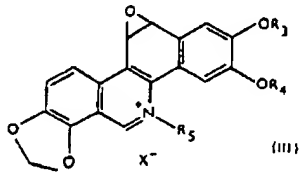
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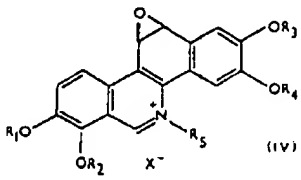
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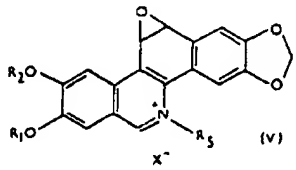
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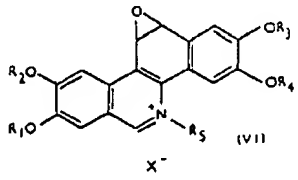
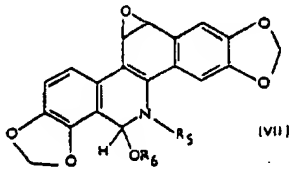

(VI)

(wherein R_1, R_2, R_3, R_4 and R_5, which may be the same or different, represent hydrogen atoms or alkyl groups, and X- represents an acid residue) either alone or in admixture, and the corresponding base forms of the above salts, of formula VII—XII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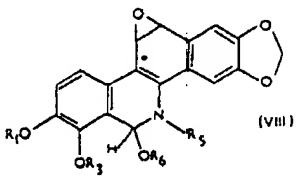
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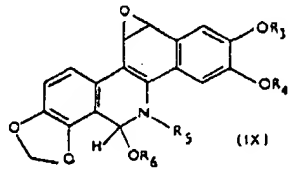
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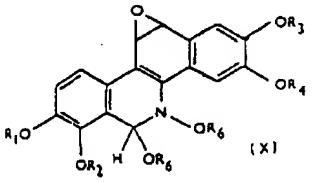
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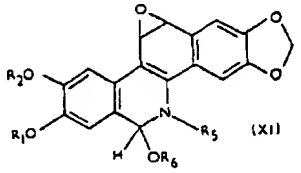
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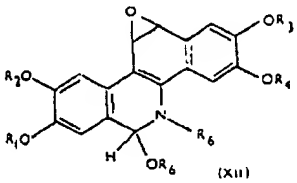

(XII)

(wherein R_1, R_2, R_3, R_4 and R_5 are as hereinbefore defined and R_6 represents a hydrogen atom or an alkyl group), either alone or in admixture.

A process for the preparation of the new compounds and pharmaceutical compositions containing them are also described.

The new compounds process valuable pharmacological activities, particularly against leukemia, bilharziasis, pseudomonas aeruginosa, benign and malignant tumours, periodontal disease and dental caries.

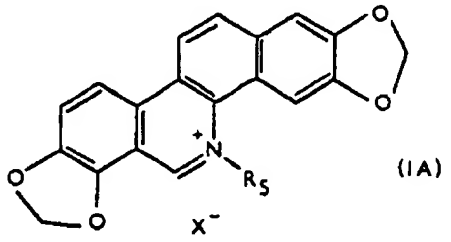
GB 2 108 955 A

This print takes account of replacement documents later filed to enable the application to comply with the formal requirements of the Patents Rules 1982.

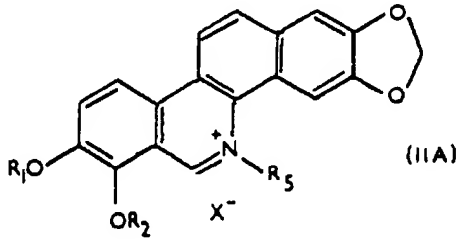
SPECIFICATION
Chemical compounds

The present invention refers to new compounds with pharmaceutical activity, namely against leukemia, bilharziasis, pseudomonas aeruginosa, benign and malignant tumours—such as granuloma pyogenicum, kerato-acanthoma, basal-cell carcinoma, squamons-cell carcinoma, malignant melanoma, kaposi's sarcoma-cell and, adeno-carcinoma of the breast—as well as periodontal disease and dental caries. This invention also refers to pharmaceutical compositions containing the said compounds and to a process for their preparation by means of a chemical reaction of epoxydation.

The cyto-static and cytotoxic activities of a type of organic compounds named benzo [c] phenanthridine alkaloids, or simply benzo [c] phenanthridines, are known. These can either be present in the form of acid salts with the structural formulae IA to VIA:



(IA)



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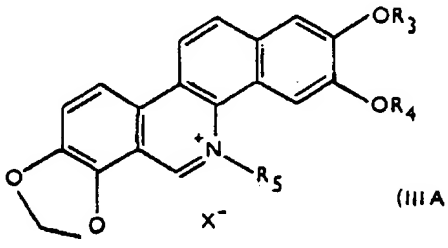

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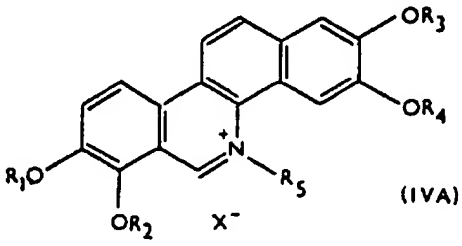

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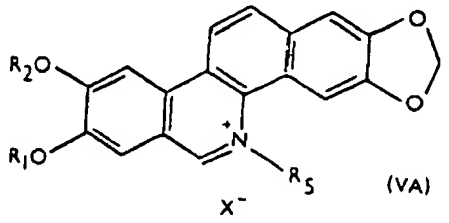
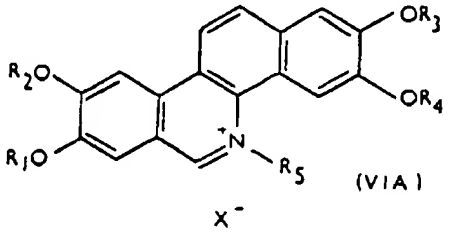
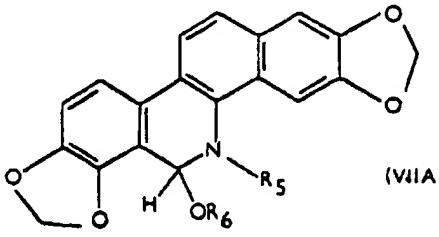


(VA)

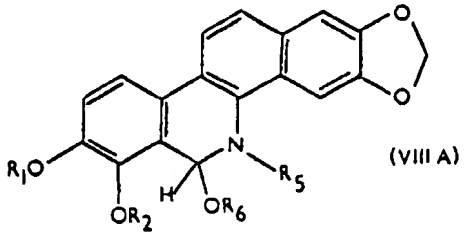


(VIA)

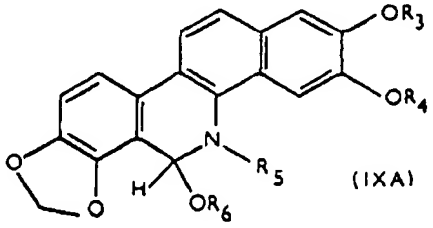
where R_1, R_2, R_3, R_4 and R_5 can be hydrogen or alkyl and X– stands, for example, for halide, nitrate, perchlorate, sulphate, hydrogen sulphate or carboxylate, or in the form of bases with the formula VIIA to XIIA:



(VIIA)



(VIII A)



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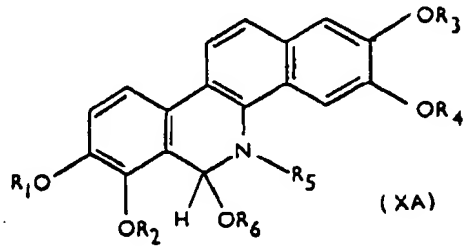
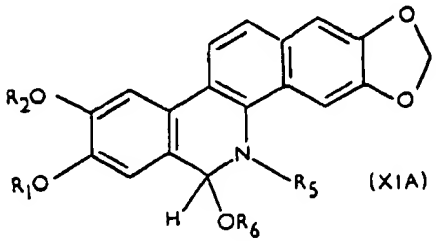
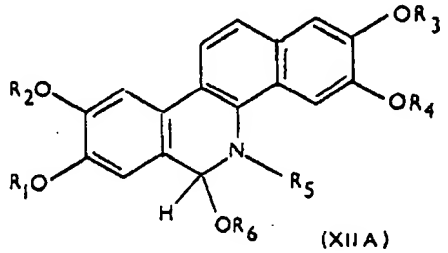


(XA)



(XIA)



(XIIA)

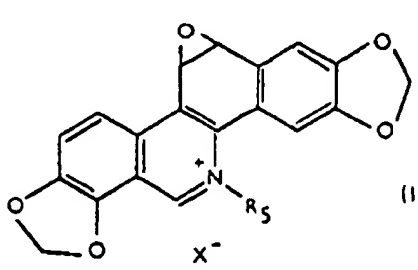
where R_1 to R_5 have the same meaning and R_6 is hydrogen or alkyl. Among these, the most important are fagaronine (formulae VIA in the salt form and XIIA in the base form, where R_1, R_2 and R_5 are methyl groups, X— is chloride, R_6 is hydrogen, and either R_3 is hydrogen and R_4 is methyl, or vice versa) considered highly cytotoxic and active against leukemia P388 in mice (W. M. Messmer, M. Tin-Wa, H. H. S. Fong, C. Bevelle, N. R. Farnsworth, D. J. Abraham and J. Trojanek, J. Pharm. Sci. *61*. 1858 (1972). and nitidine (formulae VA in the salt form and XIA in the base form, where R_1, R_2, and R_5 are methyl groups, X— is chloride and R_6 is hydrogen) equally considered highly cytotoxic and active against leukaemia P388 in mice (M. R. Wall, M. C. Wani, and H. L. Taylor, 162nd National Meeting of the American Chemical Society, Washington D.C. Sept. 1971, Abstracts MEDI 34.).

Other benzo [c] phenanthridine alkaloids such as sanguinarine (formulae IA in the salt form and VIIA in the base form, where R_6 is methyl X— is chloride and R_6 is hydrogen), also called, in its chloride salt form, 2,3:7,8-bis-methylenedioxy-5- methylphenanthridium chloride, chelerythrine (formula IIA in the salt form and VIIIA in the base form, where R_1, R_2 and R_5 are methyl groups X— is chloride and R_6 is hydrogen) also called, in its chloride salt form, 2,3-methylenodioxy-5-methyl-7,8-dimethoxyphenanthridinium chloride, which, although considered cytotoxic, are inactive against Leukaemia (F. R. Stermitz, N. A. Larson and D. K. Kim, J. Medicinal Chemistry *16* 939 (1973)).

However, there are active principles in extracts of plants, such as, for example, "sanguinaria canadensis", whose activity against cancer, in conjunction with other constituents, is claimed in the recent Portuguese patent application No. 71,727, by Orewa Inc..

In the present invention, the fact that compounds with a structure very similar to that of the benzophenanthridinic alkaloids, namely the benzophenanthrenes (the aromatic polycyclic carbohydrates with the same aromatic rings of the benzophenanthridines, differing from the latter in the replacement of the nitrogen atom by a CH group) act as inductors of cancer only through their metabolites whose formation involves the epoxydation of the double bond 11, 12 of the so called K region (D. W. Nebert, R. C. Lewit and O. Pelkonen, in "Carcinogens Identification and Mechanisms of Action" edited by A. C. Griffin and C. R. Shaw, Raven Press, New York 1979) has been taken into account. The same inductors of cancer are also known as being, simultaneously, inhibitors of carcinogenesis, which, although apparently absurd (L. W. Wattenber in the same work) can be explained by the finding that they act in different phases of the process.

According to one feature of the present invention, we provide new epoxides of benzophenanthridine alkaloids, in salt form, either alone or in admixture, with the following general formulae:



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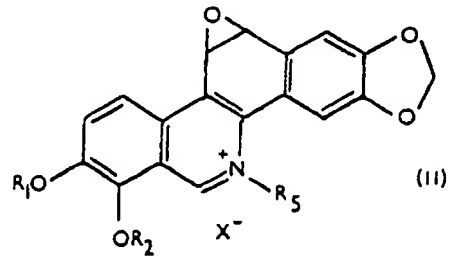

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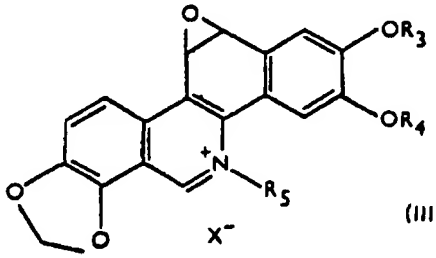

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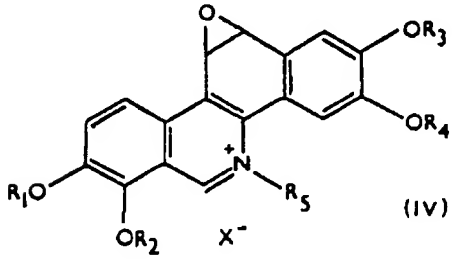

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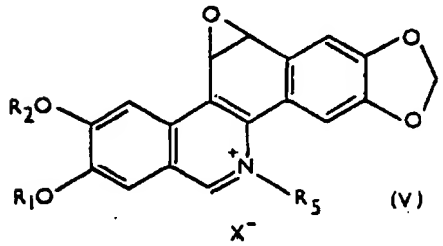

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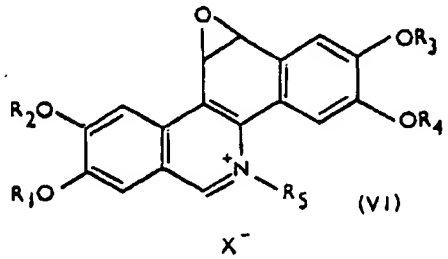
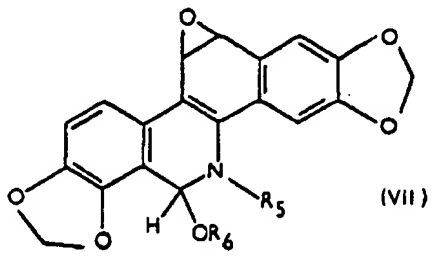


(VI)

where R_1, R_2, R_3, R_4 and R_5 can be hydrogen or alkyl and X— stands for halide, nitrate, perchlorate, sulphate, hydrogen sulphate or carboxylate or in the form of the corresponding bases, either alone or in admixture, with the following formulae:



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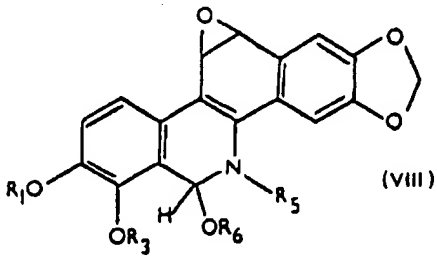

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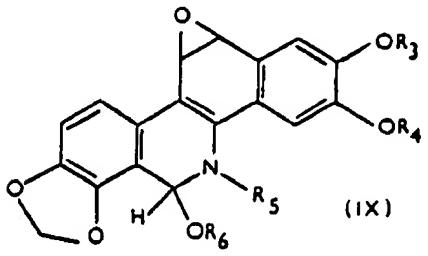

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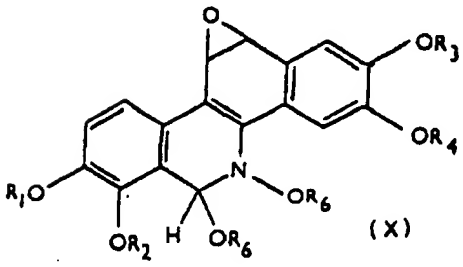

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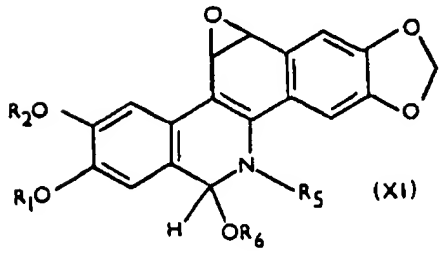

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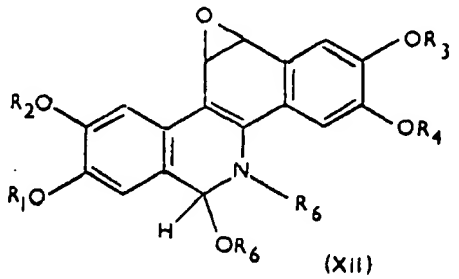
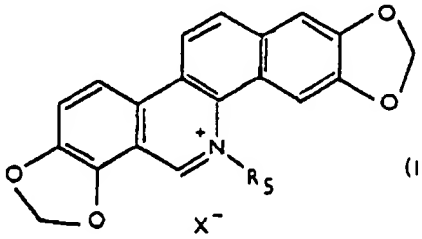


(XII)

where R_1 to R_5 have the meanings mentioned above, and R_6 is hydrogen or alkyl, which comprises epoxidation of the respective benzophenanthridine alkaloids either in the form of salt, alone or in admixture, with the following general formulae:



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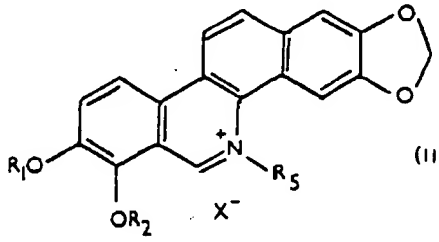

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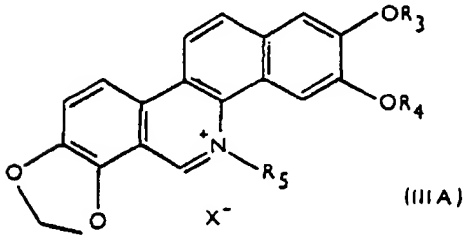

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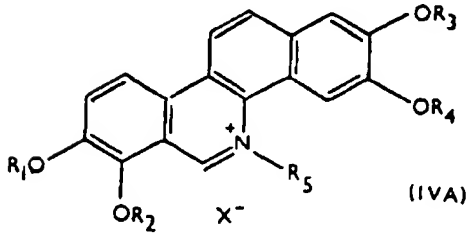

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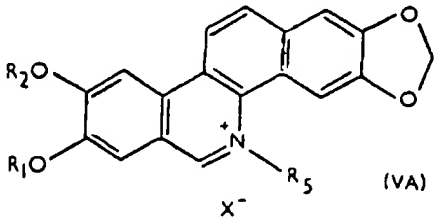
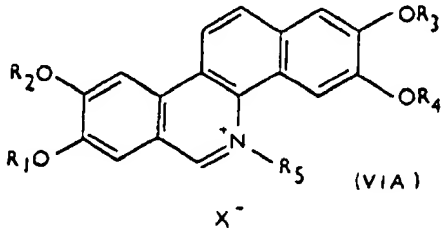
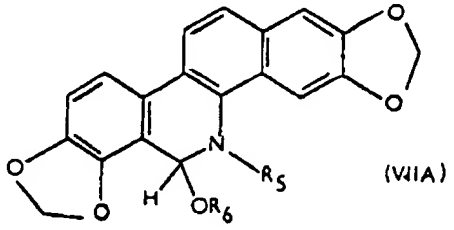


(VA)



(VIA)

where R_1 to R_5 and X− have the previously referred meanings or, in the form of the corresponding bases, either alone or in admixture, with the general formulae:



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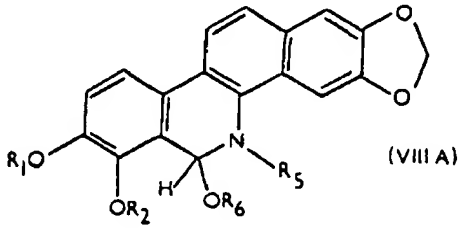

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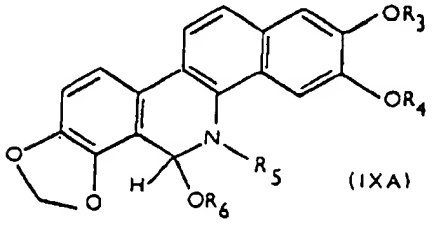

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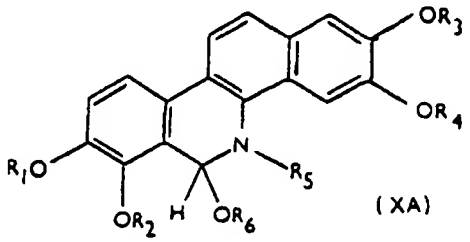

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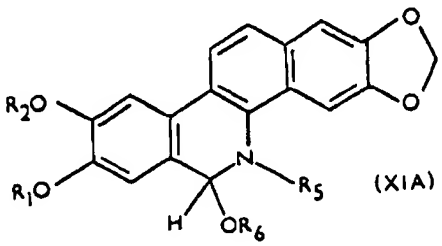
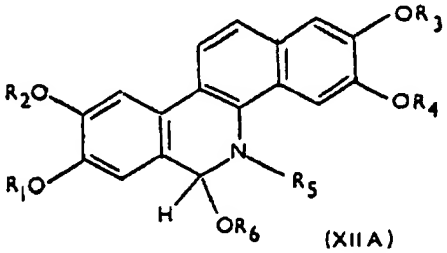


(XIA)



(XIIA)

where R_1 to R_6 have the means previously cited.

Particularly preferred compounds according to the invention are:

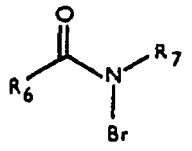
2,3:7,8-bis-methylenedioxy-5-methyl-benzophenanthridinium chloride 11,12-epoxide; and

2,3-methylenedioxy-5-methyl-8,9-dimethoxybenzophenanthridinium chloride 11,12-epoxide; and

2,3-methylenedioxy-5-methyl-7,8-dimethoxy-benzophenanthridinium chloride 11,12-epoxide

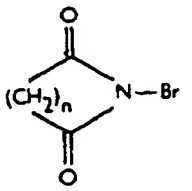
We further provide a process of synthesis of the same by means of a chain of reactions consisting essentially of two steps: a) the hydroxybromination by means of a compound containing a nitrogen-bromine bond and b) a dehydrobromination leading to the respective epoxide.

In step (a) of the process according to the invention, the hydroxybrominating agent may, for example be a compound of formula XIII



(XIII)

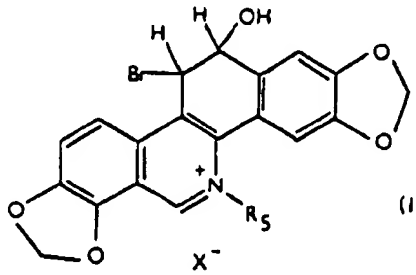
(where R_6 represents a hydrogen atom or an alkyl group and R_7 represents a hydrogen atom or an alkyl or acyl group), or a compound of formula XIV



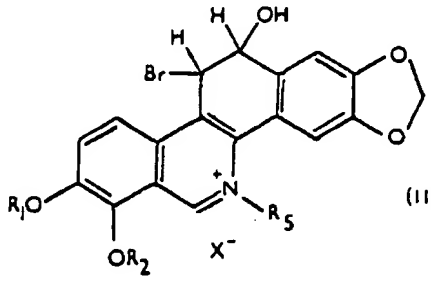
(XIV)

Preferred as hydroxybrominating agents in step (a) are 1,3-dibromo-5,5-dimethyl-hydantoin, and N-bromosuccinimide.

Step (a) is conveniently carried out in a strongly acid medium, and step (b) thus typically comprises the reaction of compounds of formulae IB—VIB



(IB)



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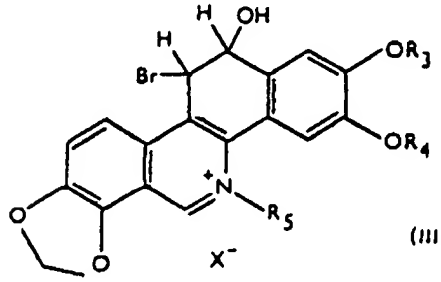

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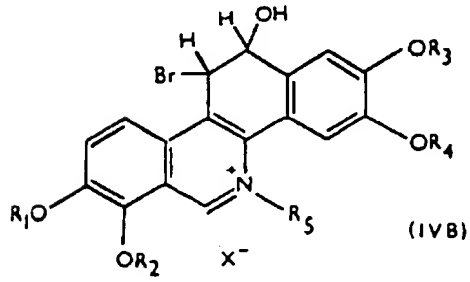
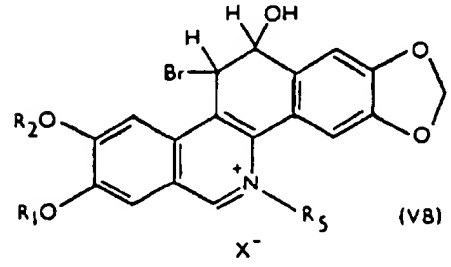
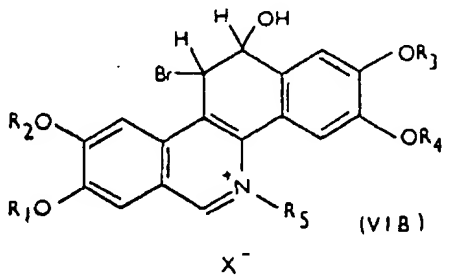


(IVB)



(VB)



(VIB)

(wherein R_1, R_2, R_3, R_4, R_5 and $X^{\ominus}$ are as hereinbefore defined, either alone or in admixture, obtained from step (a), with a strong alkali to effect elimination of hydrogen bromide and epoxide formation, to yield the compounds of the invention, either alone or in admixture.

Steps (a) and (b) may be carried out without isolation of the products of step (a).

The practical industrial interest of this process lies in new compounds with pharmaceutical activity similar to that of the basic compounds before the epoxidation is carried out, the difference being that they become more active due to the metabolic transformation in the cells where they act. Furthermore, they can be intermediates in the synthesis of other new compounds with pharmaceutical activity by opening of the epoxide ring.

The process of preparation of the epoxides may be started either from pure benzophenanthridine compounds or from the mixture of two or more, which is interesting insofar as in extracts of certain plants, such as for example sanguinaria canadensis, mixtures of two benzophenanthridine alkaloids (sanguinarine and chelery-thrine) appear, and it is more economic to epoxides the two alkaloids at the same time and use a mixture of two epoxides in the manufacture of the pharmaceutical composition. than to separate the two alkaloids and perform the epoxidations one at a time.

Finally, it should be noted that, starting from certain benzophenanthridine alkaloids as, for instance, sanguinarine, as well as from the respective epoxides, active pharmaceutical preparations are obtained only when the alkaloid, or its epoxide, is complexed by means of a metal salt. In the case of the non-epoxidated benzophenanthradine alkaloids this is a known fact, particularly with sanguinarine since 1878 (US Patent —A-209 331, of Littleton Daniel), but the application of this principle to the epoxydate benzophenanthridines is novel.

The following examples illustrate the present invention, but should not in any way limit its scope.

Example 1

Epoxidation of sanguinarine (base) (2,3:7,8-bis-methylenedioxy-6-hydroxy-5-methyl-5,6-dihydrobenzo[c]phenanthridine)

1. Hydroxybromination

2 g of sanguinarine are dissolved in 50 ml of tetrahydrofuran and stirred, and the temperature is adjusted to 20°C, 0.2 ml of perchloric acid and 1 g of dibromantin are added and stirring is required for 30 minutes. Then, 0.5 ml of a 25% sodium bisulfite aqueous solution are added. The hydroxybrominated compound formed in this reaction is not isolated, and the next reaction is then carried out.

2. Dehydrobromination

The temperature being maintained at 20°C, and under stirring, 2 g of sodium hydroxide previously dissolved in 30 ml of water are added, followed by 3 ml of acetic acid. All the tetrahydrofuran is eliminated by evaporation and the remainder is filtered, washed with water and dried in a *vacu-um* over at 40°C. 2 g of 2,3:7,8-bis-methylenedioxy-6-hydroxy-5-methyl-5,6-dihydro-benzo[c]phenanthridine 11,12-epoxide. Melting point 230.5°C UV spectrum in ethanol: λ_{max} 330, 290, 240 and 220 nm. IR spectrum (in KBr) 2940 (strong broad) 1765, 1680 (weak broad) 1640 (weak sharp) 1520 (medium sharp), 1500, 1490, 1470 (strong sharp), 1450, 1440, 1420, 1400, 1360, 1340 (weak sharp), 1250 (strong sharp), 1260, 1230, 1205, 1180 (weak sharp), 1100 (strong broad), 1005, 980 (weak sharp), 950 (medium sharp), 930, 890, 880 (weak sharp), 870 (medium sharp), 860, 850 (weak sharp), 815 (strong sharp), 790, 770, 760, 720, 700, 680, 625 (weak sharp), 470 (strong board). NMR spectrum (300 MHz, d_6 MDSO TSS): δ 2.759 (>N—CH_3), δ 3.225, 3.372 ($H_{11;12}$) δ 5.406 (H_6) δ 6.200, 6.259, 6.298 (2×O—CH —O, 2 doublets respectively centred at 6.249 and 6.278) 7.112, 7.136, 7.617, 7.641 ($H_{1,4}$ AB system centred at 7.375; J_{ab}=—7.3 Hz), 7.504, 5.734, 8.264 ($H_{9,10}$) are obtained.

Example II

Transformation of sanguinarine (base epoxide, that is 2,3:7,8-bis-methylene-dioxy-6-hydroxy-5-methyl-5,6-dihydrobenzo[c]phenanthridine 11,12-epoxide into the respective chloride of the sanguinarinium epoxide, that is 2,3:7,8-bis-methylene-dioxy-6-hydroxy-5-methyl-5,6-dihydrobenzo[c]phenanthridinium chloride 11,12-epoxide.

1 g of the base is dissolved in chloroform or acetone and aqueous hydrochloric acid is then added, drop by drop, until a substance of a bright orange colour is precipitated; this substance is separated by filtration, washed with chloroform and dried in vacuum at room temperature. Melting point between 300 and 350°C, with decompostion.

Example III

Epoxidation of chelerythrine (base) (2,3-methylenedioxy-6-hydroxy-5-methyl-7,8-dimethoxy-5,6-dihydrobenzo[c]phenanthridine).

The procedure is quite similar to that of Example 1, the difference being in starting from 2 g of chelerythrine (base), instead of sanguinarine (base), thus obtaining 2 g of chelerythrine epoxide, that is, of 2,3-methylene-dioxy-6-hydroxy-5-methyl-7,8-dimethoxy-5,6-dihydrobenzo[c]phenanthridine 11,12-epoxide.

Melting point 250.0°C. UV spectrum in ethanol: λ_{max} 340, 285, 230 and 210 nm. IR spectrum (in KBr): 2940 (strong Broad), 1765 (weak broad) 1680 (medium sharp), 1635, 1620, 1565 (weak broad) 1520 (weak sharp), 1500 (strong sharp) 1465, 1430, 1400, 1370, 1370, 1330, 1310 (weak sharp) 1275 (strong sharp) 1220, 1190, 1120 (weak broad), 1050 (strong sharp) 1000 (weak broad), 955 (medium sharp), 915, 900 (weak broad), 870 (medium sharp), 840 (weak broad), 815 (medium sharp), 730, 675, 605, 525 (weak broad) and 475 (strong broad). NMR spectrum (300 MHz, d_6—DMSO-TSS): δ 0.868, 1.245, 2.725, 3.176, 3.191, 3.745, 3.779, 3.848, 3.901, 4.191, 5.357, 5.646, 5.798, 5.833, 5.847, 6.127, 6.151, 6.181, 6.279, 6.303, 7.033, 7.063, 7.347, 7.449, 7.548, 7.582, 7.783, 7.857, 7.881, 7.851.

Example IV

Preparation of a mixture of sanguinarine (base) and chelerythrine (base) epoxydes starting from an extract of "sanguinaria canadensis" containing a mixture of sanguinarine (base) and chelerythrine (base).

100 g of dried and ground rhizomes of "sanguinaria canadensis" are extracted with 2 portions of 500 ml of methyl alcohol; the solution is filtered through an alumina column, rejecting the first fractions, and the methyl alcohol is evaporated until abundant precipitation of crystals occurs, and these are separated by dry filtration, 2 g of extract are obtained and, by thin layer chromatography on silicagel (10% methanol and 90% methylene chloride) sanguinarine is shown as the main constituent followed by chelerythrine. The 2 g of extract are submitted to the operations described in Example 1. A product is obtained which, by thin layer chromatography and by comparison with the sanguinarine and chelerythrine epoxides proves to be a mixture in which the sanguinarine epoxide prevails over the chelerythrine epoxide.

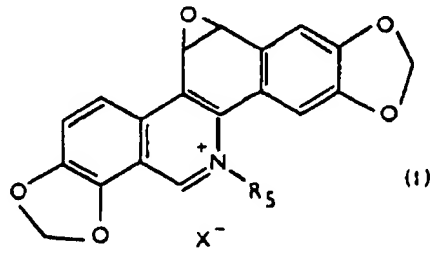
Example V

Preparation of a pharmaceutical composition

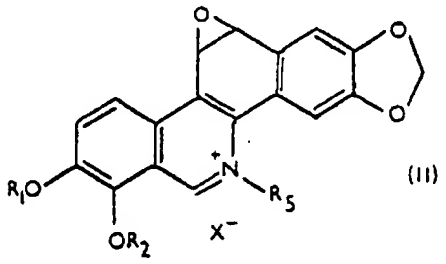
A paste is prepared with 45 g of zinc chloride, 37 ml of deionized water, 16 g of sanguinarine and 2 g of sanguinarine epoxide.

Claims

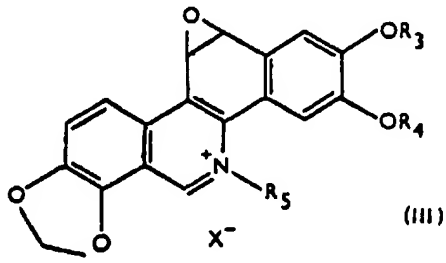
1. Process for the preparation of new epoxides of benzophenanthridine alkaloids, in salt form, either alone or in admixture with the following general formulae:



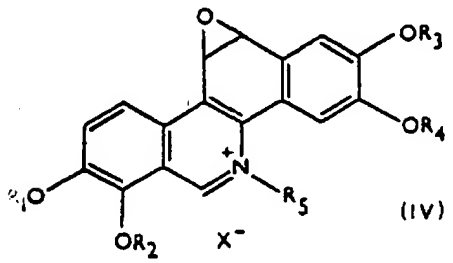
(I)



(II)



(III)



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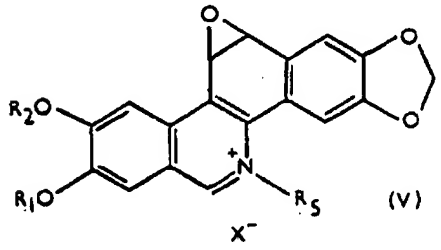
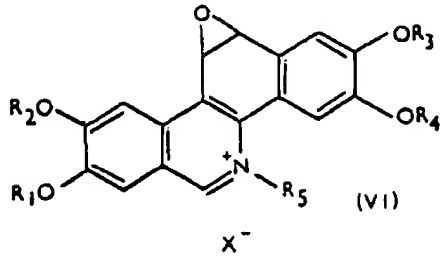
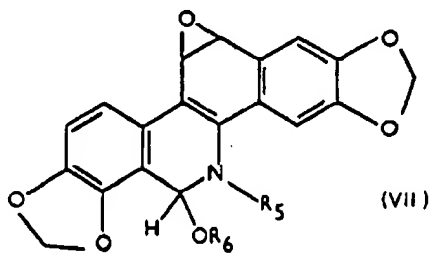


(V)

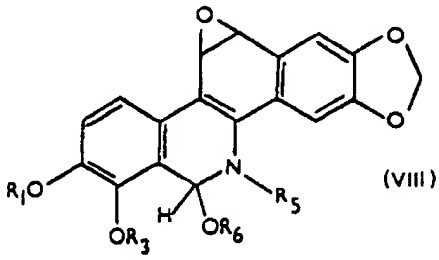


(VI)

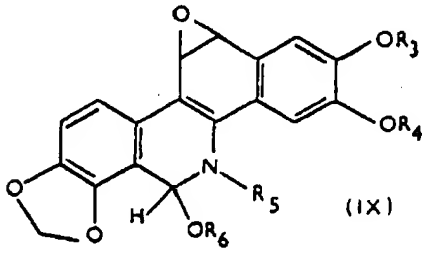
where R_1, R_2, R_3, R_4 and R_5 can be hydrogen or alkyl and X— stands for halide, nitrate, perchlorate, sulphate, hydrogen sulphate or carboxylate or in the form of the corresponding bases, either alone or in admixture, with the following formula:



(VII)



(VIII)



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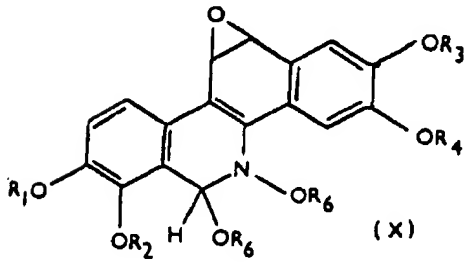
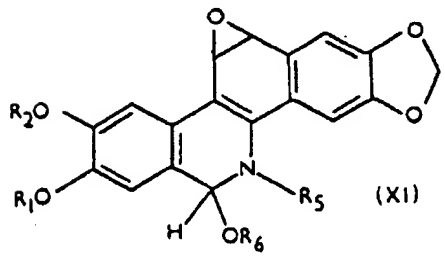
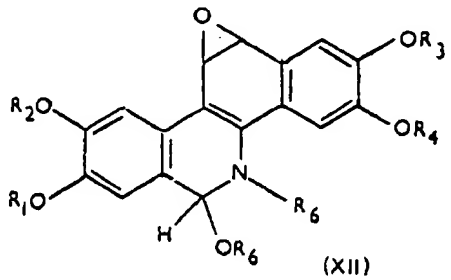


(X)

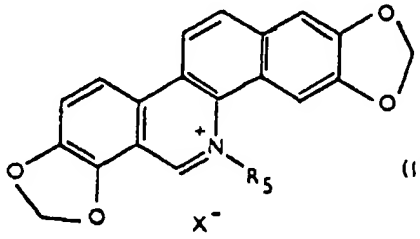


(XI)

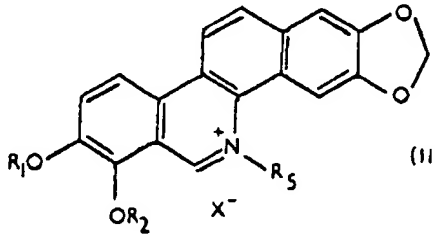


(XII)

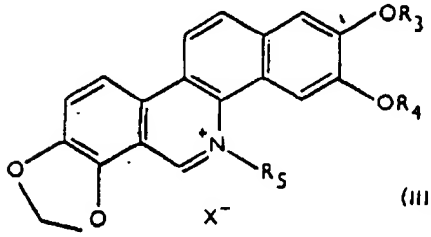
where R_1 to R_5 have the meanings mentioned above, and R_6 is hydrogen or alkyl, which comprises epoxidation of the respective benzophenanthridine alkaloids in the form of salt, alone or in admixture, with the following general formulae:



(IA)



(IIA)



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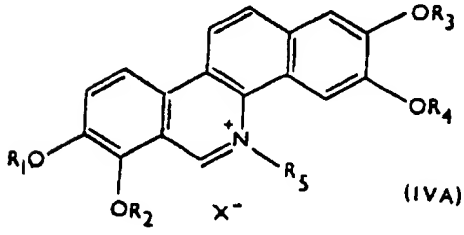
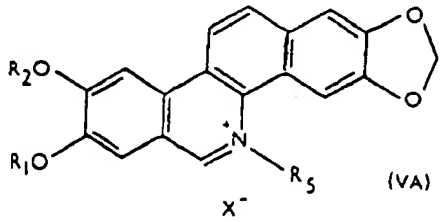
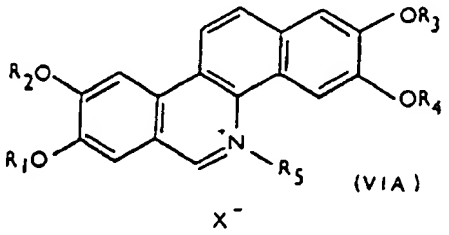


(IVA)

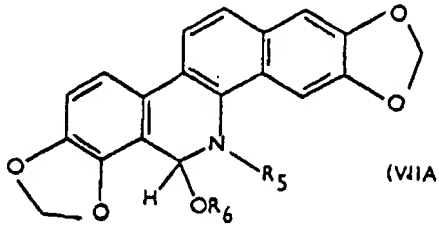


(VA)

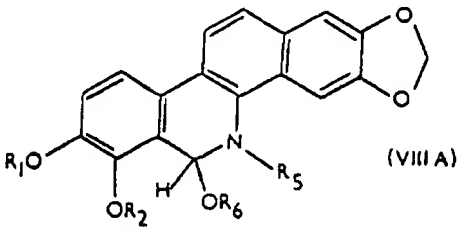


(VIA)

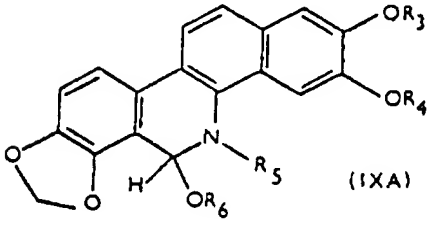
where R_1 to R_5 and X— have the previously referred meanings or in the form of the corresponding bases, either alone or in admixture, with the general formulae:



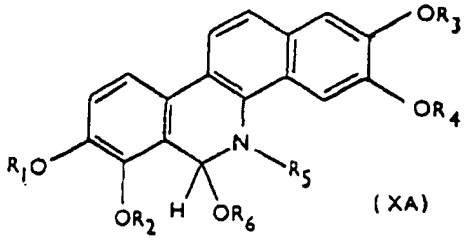
(VIIA)



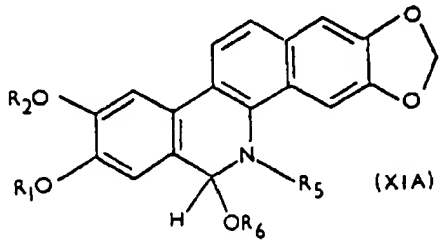
(VIII A)



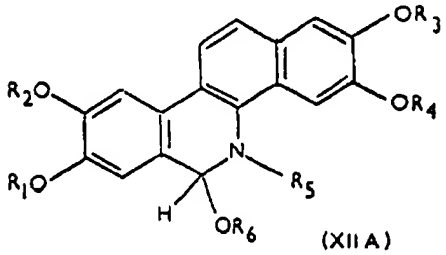
(IXA)



(XA)



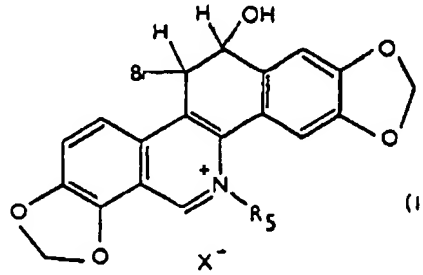
(XIA)



(XIIA)

where R_1 to R_6 have the meanings previously cited.

2. Process according to Claim 1, in which the epoxidation is performed by means of a strongly acid medium reaction of benzophenanthridine alkaloids with a compound with a nitrogen bromine bond, thus obtaining the respective hydroxybrominated derivatives, either alone or in admixture, with the following general formulas:



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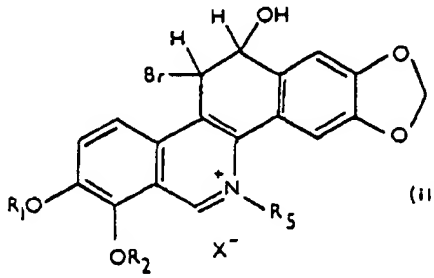
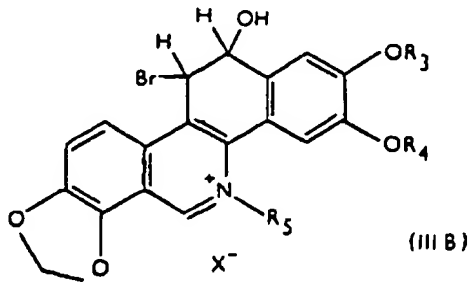
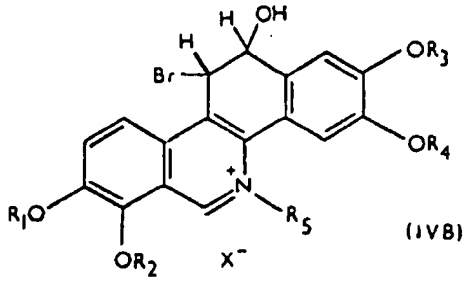


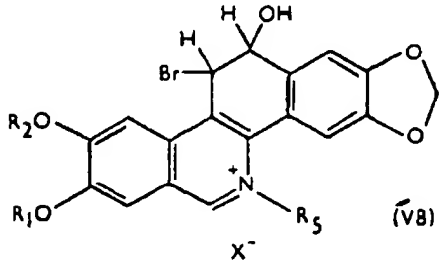
(IIB)



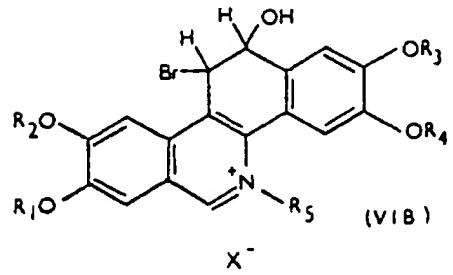
(III B)



(IV B)



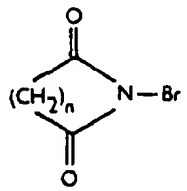
(V B)



(VI B)

where R_1 to R_8 and X– have the meanings previously cited and, by subsequently reacting with a strong alkali, undergoing the elimination of hydrogen bromide in order to form the respective epoxides, either alone or in their mixtures.

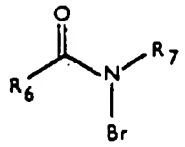
3. Process according to Claim 2, in which the compound containing the N-bromine bond has the general formula:



(XIV)

where the R_8 group may be hydrogen or alkyl and R_7 may be hydrogen, alkyl or acyl.

4. Process according to Claim 2, in which the compound containing the nitrogen bromine bond as the general formula:



(XIII)

where n is an integer.

5. Process according to Claim 2, in which the compound containing the nitrogen bromine bond is 1,3-dibromo-5,5-dimethyl-hydantoin or dibromatinone.

6. Process according to Claims 1 to 5, in which one or more of the following compounds are obtained: 2,3:7,8-bis-methylene-dioxy-5-benzophenanthridinium chloride 11,12-epoxide; 2,3-methylene-dioxy-5-methyl-8,9-dimethoxybenzophenanthridinium chloride 11,12-epoxide; methylene-dioxy-5-methyl-7,8-dimethoxy-benzophenanthridinium chloride 11,12-epoxide.

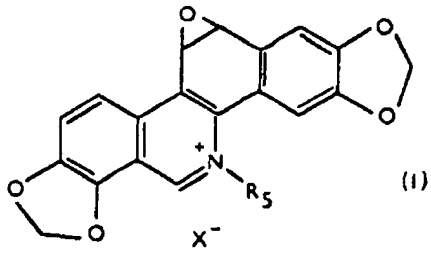
7. Process for the preparation of pharmaceutical compositions having as active ingredients an epoxide of a benzophenanthridinic alkaloid with the formulas (I) to (XII) as claimed in claim 11 or a mixture of several of those epoxides of benzophenanthridine alkaloids, jointly with one or more non-epoxidated benzophenanthridine alkaloids or not, with the formulas (IA) to (XIIA) combined with one or more vehicles pharmaceutically acceptable and adequate for therapeutic administration.

8. Process for the preparation of pharmaceutical compositions according to Claim 7, in which the benzophenanthridine alkaloids and the respective epoxides incorporated in the composition are activated by complexation with a metal.

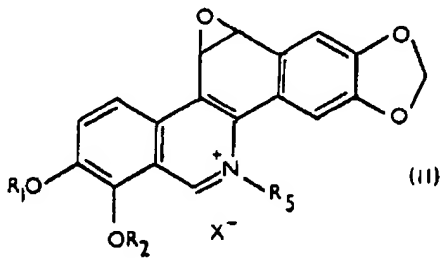
9. A process as claimed in any preceding claim substantially as herein described.

10. A process as claimed in any preceding claim substantially as herein described in any of Examples I to V.

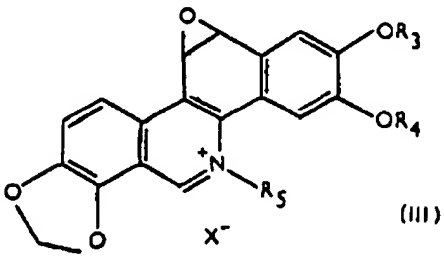
11. Compounds of formulae I—VI



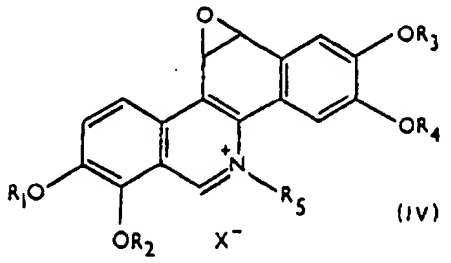
(I)



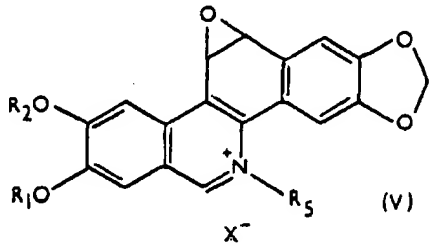
(II)



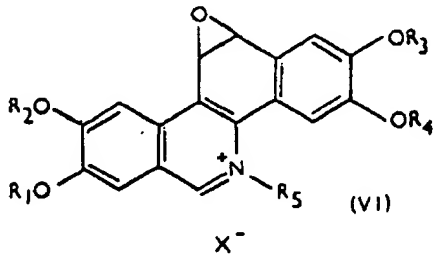
(III)



(IV)

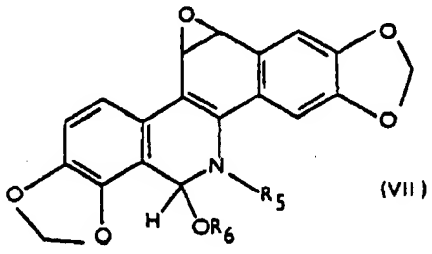


(V)

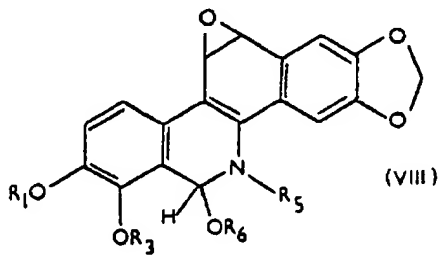


(VI)

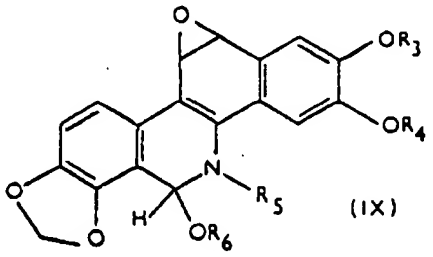
(wherein R_1, R_2, R_3, R_4 and R_5, which may be the same or different, represent hydrogen atoms or alkyl groups, and X— represents an acid residue) either alone or in admixture, and the corresponding base forms of the above salts, of formulae VII—XII



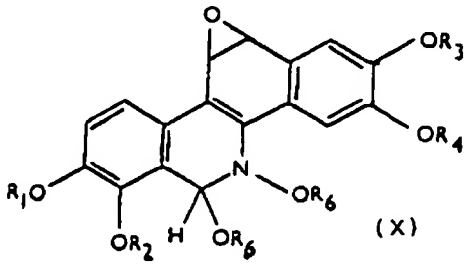
(VII)



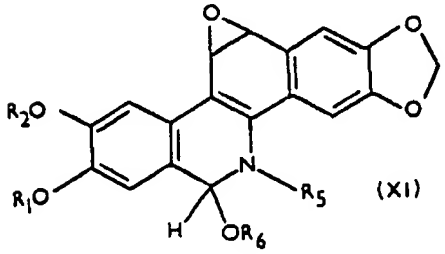
(VIII)



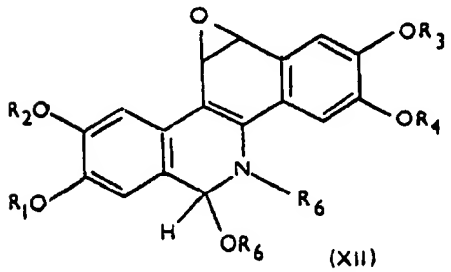
(IX)



(X)



(XI)



(XII)

(wherein R_1, R_2, R_3, R_4 and R_5 are as hereinbefore defined and R_6 represents a hydrogen atom or an alky group), either alone or in admixture.

12. Compounds as claimed in claim 11 wherein X— represents a halide, nitrate, perchlorate, sulphate, hydrogen sulphate or carboxylate ion.

13. 2,3:7,8-bis-methylenedioxy-5-methyl-benzophenanthridinium chloride 11,12-epoxide.

14. 2,3-methylenedioxy-5-methyl-8,9-dimethoxybenzophenanthridinium chloride 11,12-epoxide.

15. 2,3-methylenedioxy-5-methyl-7,8-dimethoxybenzophenanthridinium chloride 11,12-epoxide

16. Compounds as claimed in any one of claims 11 to 15 claim as herein described.

17. Compounds as claimed in any one of claims 11 to 16 claim as herein specifically described in any one of Example I to IV.

18. Pharmaceutical compositions comprising as active ingredient at least one compound as claimed in claim 11, optionally together with at least one non-epoxidised benzophenanthridine

alkaloids, of formulae IA to XIIA as defined in claim 1, in association with one or more pharmaceutical carriers or excipients.

19. Pharmaceutical compositions as claimed in claim 18 wherein the active ingredient(s) is/are activated by complex formation with a metal.

20. Pharmaceutical compositions substantially as herein described.

21. Pharmaceutical compositions substantially as herein described in Example V.

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